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## A NEW METHOD OF MOUNTING SUPERFICIAL FUNGI.

HERBERT H. WHETZEL.

In Studying *Sphaeropsis malorum* on apple leaves, I found that in its early stages, before pycnidia were formed, well defined brown spots appeared. Although I examined many sections from these brown spots, I failed to discover any mycelium. Material scraped from the surface showed nothing of the mycelial character of the fungus. I had about decided that the spots were not due to a fungus when I found a few immature pycnidia on one of the spots. It then occurred to me to cook a piece of the leaf in a weak solution of Potassium hydrate (KOH) and by macerating the leaf tissues thus to discover the mycelium. I had successfully used this method in studying mycelium distribution in the galls of *Gymnosporangium macropus*. After slowly cooking the leaves for half an hour, I found that the brown coloring of the leaves was bleached out. The epidermis separated as a thin sheet from the other tissues of the leaf. After washing thoroughly in clear water, I examined it under the microscope and found, that, while the leaf tissue was entirely bleached, the brown threads of the fungus still retained their color and showed beautifully.

The same method used on several other fungi growing superficially on leaves, stems and fruits has given in every case excellent results. This method has never before been used, so far as I know, and believing that other persons may find it helpful, I give it in detail as I have worked it out.

1. Carefully peel or slice off a piece of the epidermis on which the fungus is growing. Care should be exercised to take away as little subepidermal tissue as possible.

2. Immerse the slice in a 2-4% solution of Potassium hydrate and boil in an evaporating dish over a low flame for 20-30 min. Cook long enough to remove all the color from the tissue of the host.

3. Pour off the Potassium hydrate solution and wash by letting the material stand for 10-20 min. in each of two or three changes of clear water, stirring it about occasionally. If all the color is not removed from the host tissue cook again. Carefully pick away any pieces of subepidermal tissue that may cling to the epidermis.

4. Dehydrate in 95% alcohol.

5. Clear in carbol clearer (2 parts pure carbolic acid and 3 parts turpentine.)

6. Mount in Canada balsam.

The Advantages of this method may be summed up as follows:

1. The mycelium, pycnidia, etc., are preserved in their natural form and position on the surface of the host, and not all broken and disarranged as is the case when they are scraped from the surface.

2. In many cases of the forms the mycelium is so scanty that in an ordinary mount or section too little is present to show its nature, while by this method a large area is presented so that all the mycelium is sure to be present.

3. Pycnidia in all stages of development may be easily obtained and studied in their normal position and relation to the mycelium on which they are borne.

4. In order that the spores may be shown, a pycnidium at the side of the mount may be crushed with a dry scalpel while the material is still in the clearer.

5. By removing the color of the host cells there is nothing to interfere with the examination of the mycelium by transmitted light.

This method will be found most valuable in the case of those fungi having dark colored mycelium. Hyaline mycelium would be scarcely more evident than the colorless host cells. The whole secret of the process lies in the fact that the pigment in most of the higher plants (the hosts) is bleached by Potassium hydrate, while that of the parasite is not affected.

Very excellent results have been obtained in the case of the following fungi: Cladosporium carpophilum on the bark of peach twigs, Leptothyrium pomi on the fruit of cultivated apples, Vermicularia circinans on the outer leaves of onion bulbs, Sphaeropsis malorum on leaves of cultivated apples, Macrosporium cumerinum on leaves of Cucumis melo.

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## THE FINDING OF PUCCINIA PHRAGMITIS (SCHUM.) KORN. IN NEBRASKA.

BY JOHN M. BATES.

On June 14th, 1901, while collecting asparagus rust in a garden in Kearney, Nebr., I found six or seven spots of pure white aecidia on *Rheum raphonticum* on the under side of the leaves. It was at once pronounced to be *Puccinia phragmitis*, and appeared to be on a new host for this country. This suggested further study of the species. This garden is three miles from the Platte River, and probably there is no patch of *Phragmites phragmitis* nearer than this. About a mile west of Callaway, Custer County, there are several patches of this grass, and on this I found rust on the 28th of August of last year. The Uredo stage was just passing away at that date. This brought